EXHIBIT A

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant:	Dorit PLAT et al.	Confirmation No. 3410
Serial No.:	10/572,782	Examiner: Abigail L. FISHER
Filed:	November 8, 2006	Art Unit: 1616
For:	STABILIZED FORMULATIONS OF PHOSPHATIDYLSERINE	

DECLARATION OF Gai Ben-Dror
UNDER 37 C.F.R. § 1.132

In the Matter of
US Patent Application Ser. No. 10/572,782
Stabilized Formulations of Phosphatidylserine (hereafter "**the patent application**")

I, Gai Ben-Dror, hereby declare and state as follows:

I am a Chemical and Biotechnology engineer since the year 1996. I have been employed by Enzymotec Ltd. since the year 2002, and am currently employed as the Director of Process Development of Enzymotec Ltd. My CV is attached hereto (**Annex A**).

I am a co-inventor of the invention subject of the above-identified application.

I have read and understand the patent application. I have also read and understand the amended claims, attached hereto as **Annex B**, to be filed with this declaration and agree that the claims express our invention. I have also read and understood EP 922707, hereafter De Ferra. I have also read the declaration by Ms. Neta Scheinman concurrently filed with my declaration.

The oil-insoluble divalent PS salt in powder form is prepared in a substantially aqueous (monophasic) system, as shown, e.g., in the example given under 1. A (page 2 of the declaration by Ms. Neta Scheinman). This system employs non-solubilized lecithin as the starting material for making the PS, and does not contain any significant amounts of organic solvents. This preparation system is thus characterized by being substantially aqueous, and by employing non-solubilized lecithin as the substrate of the enzymatic reaction between the lecithin and the added serine. Since lecithin is rather sticky, the reaction of lecithin with the serine may be facilitated by the addition of small quantities of organic solvent before initiating the enzymatic reaction. This feature is optional, and may be used for improving the contact between lecithin and serine and assist the dispersion of the lecithin in the reaction mixture, without solubilizing (dissolving) the lecithin in the reaction system. This option of adding a small amount of an organic solvent to the reaction system before the addition of lecithin is mentioned in Example 1 of the patent application as strictly an optional feature. This option is absent, for example, from the synthetic procedure described in the Scheinman declaration.

The following experiments were conducted at my request and under my direct supervision and control, and demonstrate that PS prepared in a diphasic reaction system as taught by the prior art (De Ferra), in which the lecithin is dissolved in the organic phase before the reaction with the serine is initiated, is soluble in oils, for example edible MCT as shown below (Example 1, Solubility Test). When the PS produced by this method of the prior art was incorporated into an oil base (edible MCT), it formed a clear solution and not a dispersion.

In contradistinction, in the present invention the PS is prepared in a substantially aqueous system and the starting material lecithin is not solubilized in the reaction mixture before initiation of the reaction with the serine. It was surprisingly found that PS divalent metal salts prepared by this method, from non-solubilized lecithin, cannot be dissolved in oils, and are thus insoluble in oils, including edible oils. This is demonstrated in the example 1.A of the Scheinman declaration in which the PS calcium salts was produced in a substantially aqueous system, from non-solubilized lecithin. As

mentioned in paragraphs 7 and 8 of the example, when the resulting PS was mixed with MCT, it did not dissolve, but formed a dispersion in the oil.

Capsules containing a dispersion of PS divalent salt produced in a substantially aqueous system from non-solubilized lecithin, surprisingly exhibit prolonged storage stability, as shown in the specification, and comparatively shown in the Scheinman declaration, in contrast to capsules containing PS prepared by the process of the prior art.

The prolonged storage stability is extremely important in ensuring the consumer of a content of an effective dose in the dosage form.

I participated in the design of and approved all experiments. I continuously monitored the experiments in order to assure that they would be carried out according to their design.

I. Repeating Example 2 of De Ferra et al. (EP 0 922 707), and demonstrating that the resulting PS is soluble in oil.

Preparation of organic phase:

1. 200 g of lecithin (Epikuron 135F, Cargill, Germany),1500 ml toluene and 50 ml water were introduced into a 3000 ml reactor, under nitrogen and the mixture was concentrated under vacuum at 45^{0}C, distilling about 1000ml of the solvents. A clear solution of toluene containing the solubilized lecithin was obtained.

Preparation of aqueous phase:

Another 2000 ml Erlenmeyer was loaded with 11 g calcium chloride, 14 g sodium acetate trihydrate, 700 ml distilled water, 8.5 g acetic acid, 300 g L-serine (Kyowa, Japan) and 9.6 g PLD, that were mixed until dissolved (pH= 4.2). A clear solution was obtained.

Diphasic reaction:

1. The two clear solutions were combined and the resulting mixture was heated to and kept at 25-40^{0}C with strong stirring for 4 days.
2. HPTLC analysis showed a PS content of 85% of the total phospholipids.
3. The mixture was then added to a suspension of 13 g decalite in 250 ml toluene and filtered, washing the filter with 200 ml of toluene/water (3/1, V/V).
4. The aqueous phase was separated from the organic phase. The organic phase, after further filtration on decalite, was concentrated under vacuum to an about 200 g residue.
5. 120 g of the residue were taken up into 1300 ml acetone and stirred for 6 hr at room temperature.
8. After cooling the mixture to 4^{0}C, the product was filtered and dried under vacuum to give about 55 g of PS in the form of calcium salt.

Solubility test

23 g of the resulting PS (calcium salt) were dissolved in 24 g edible MCT, to form a clear solution.

Further testing was undertaken, which demonstrates that producing PS in a diphasic reaction results in a soluble PS:

II. Preparation of fluid PS from soy lecithin:

Preparation of organic phase:

1. 1020 g vegetable lecithin containing 20% PC, and 6800 ml ethanol were incubated in Erlenmeyer flasks at 20-25^0 C for 2 hr with shaking at 200-250 rpm.
2. The Erlenmeyer content was filtered through Whatman No.41 paper and the extract was concentrated under vacuum evaporation until reaching a honey-like texture.
3. Hexane and vegetable oil were added to the extract and the solvent mixture was evaporated under vacuum until no solvent residue was seen.
4. 500 ml hexane were added twice and evaporated under vacuum, and the resulting lecithin/vegetable oil were dissolved in a mixture containing hexane, to give a clear solution of lecithin.

Preparation of aqueous phase:

1. 11.25 g calcium chloride and 2.11 g acetic acid were added to 450 ml distilled water, titrated with caustic soda to desired pH, and heated to 40^0C.
2. 150 g L-serine and 3 g PLD were added and mixed until fully dissolved.

Diphasic reaction:

1. The clear organic phase was added to the aqueous phase.
2. The resulting diphasic system was stirred for about 48 h at 40^0C under nitrogen environment.
3. The organic phase was separated from the aqueous phase, filtered and concentrated under vacuum and evaporation was continued until full removal of the solvent to give 132 g **clear** fluid oil containing 25% soluble PS calcium salt.

It is my understanding and belief that the differences between the physical and chemical characterization of the phosphatidylserine (PS) of the subject application and the PS described in De Ferra *et al.* (EP 922707) may be explained as follows:

Although the phosphatidylserines (PS's) in both cases are in the form of their salt with a divalent ion, their solubility characteristics are different. The difference in solubility characteristics is believed to result from the fact that PS can arrange in different forms of molecule groups, according to the reaction medium in which it was produced.

In De Ferra *et al.*, during the reaction, the lecithin starting material is dissolved in a non-polar organic solvent, and it is believed that the phospholipids tend to form micelles where their non-polar side (the fatty acids "tails") turn toward the surrounding medium (the non-polar organic solvent in this case).

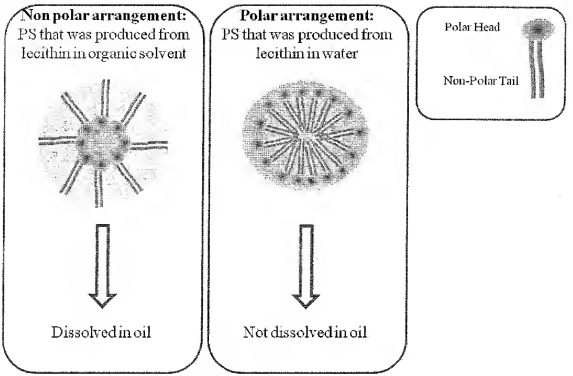
In contrast, in the subject invention, during the mono-phasic reaction in water, which is a polar medium, the lecithin is not solubilized and it is believed that the phospholipids

form the opposite arrangement, and turn their polar head group (the "phosphoserine") outside, toward the medium (which is water, i.e. polar), while their fatty acid tails (non-polar) turn away from the medium. Those two forms of arrangement of the molecules in the formed PS are believed to be retained after the PS is isolated.

This explanation is believed to clarify the different solubility characteristics of PS in oil, which is a non-polar medium. PS that was produced from lecithin in an organic solvent, by bi-phasic reaction, has a form of a "non-polar arrangement", where the fatty acids tails are turned to the outer side of the micelles, i.e. toward the medium. When PS with a "non-polar arrangement" is mixed with oil, which is non-polar, it will be dissolved in the oil and thus form a solution. This is the case with De Ferra *et al.*

On the other hand, a PS that was produced from lecithin in a water system, by mono-phasic aqueous reaction, has a form of a "polar arrangement", where the fatty acids tails are turned to the inner side of the micelles, and the polar side of the PS is turned outwards, i.e. toward the medium. When PS with a "polar arrangement" is mixed with oil, which is non-polar, it will be "rejected" by the non-polar oil and therefore will not be soluble in the oil. This PS "polar arrangement" will form a dispersion, rather than a solution in oil. This is the case with the PS of the present invention.

This can be illustrated as follows:



As shown in the Scheinman Declaration, the PS divalent salt which is dispersed in the oil, rather than dissolved in the oil, possesses superior storage stability.

Summary

1. To obtain a divalent salt of PS, the prior art only discloses the preparation of PS calcium salt in a diphasic system, in a process in which the starting material lecithin is dissolved in an organic solvent before initiation of the enzymatic reaction with serine. PS calcium salt produced according to the prior art is soluble in an edible oil [see above, Example 1 which repeated the process of Example 2 of De Ferra, and the additional Example 2].

2. The PS divalent metal salt of the present invention is prepared in a substantially aqueous system, by enzymatically reacting serine with lecithin in the presence of a

phospholipase, in which the starting material lecithin is not solubilized in the reaction system before initiation of the enzymatic reaction with serine. Unexpectedly, the PS divalent salt produced by this aqueous method is insoluble in an edible oil. When the PS divalent salt is mixed with an edible oil, it does not dissolve in the edible oil but remains in the solid state and only disperses [see the Scheinman declaration, Example 1, paragraph 7].

3. As shown in the Scheinman declaration, the dispersion in edible oil of the PS divalent salt prepared in a substantially aqueous system from lecithin which is not solubilized in the reaction system, results in solid phase particles of the PS divalent salt dispersed in edible oil that exhibits superior storage stability compared to commercial PS preparations, made from known PS salts.

The undersigned declares further that all statements made herein of her own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application of any patent issuing from the application referenced herein.

Date: 17/02/2011 By: _____

Gai Ben-Dror